

PRODUCTION OF GP88 BY TUMORIGENIC AND NON-TUMORIGENIC CELLS



FIG.1 A

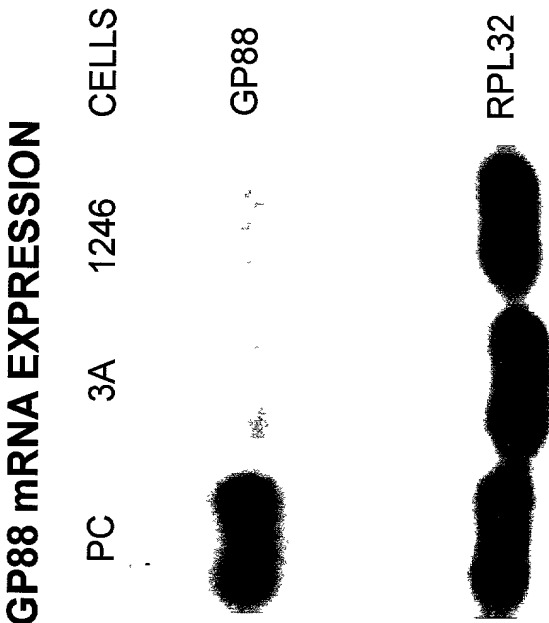


FIG.1B

GP88 mRNA EXPRESSION IN VARIOUS CULTURE CONDITIONS

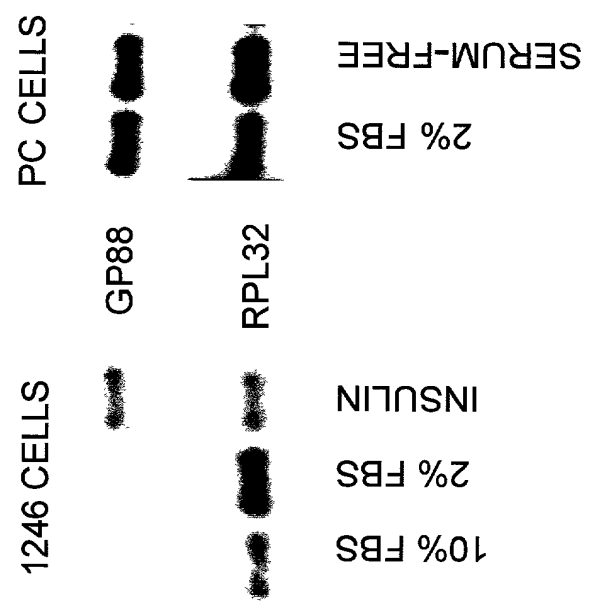


FIG.1C

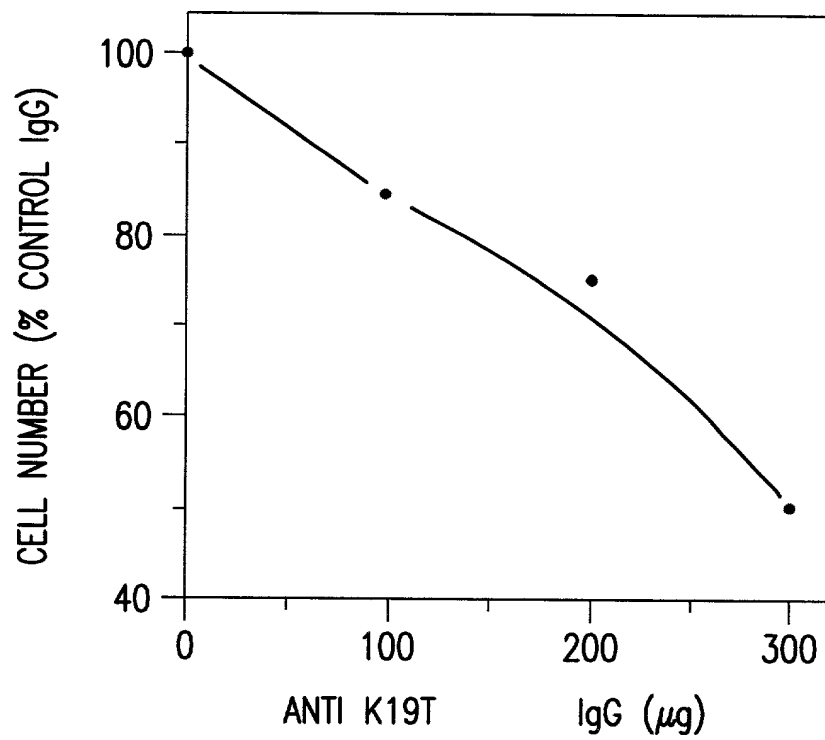
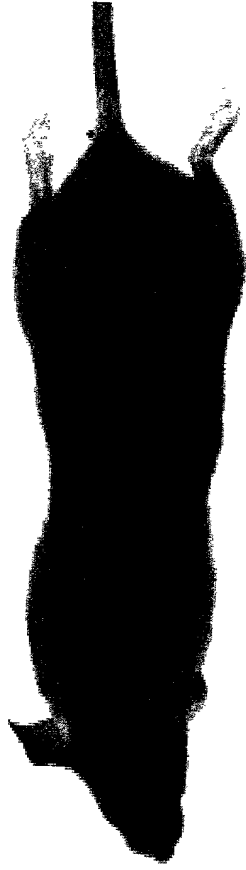


FIG.2

**ABSENCE OF TUMOR FORMATION IN C3H MICE BY INHIBITION OF GP88
EXPRESSION**



GP88 ANTISENSE TRANSFECTED PC CELLS



CONTROL TRANSFECTED PC CELLS

FIG.3

GP88 PROTEIN EXPRESSION IN TUMOR AND SURROUNDING TISSUES

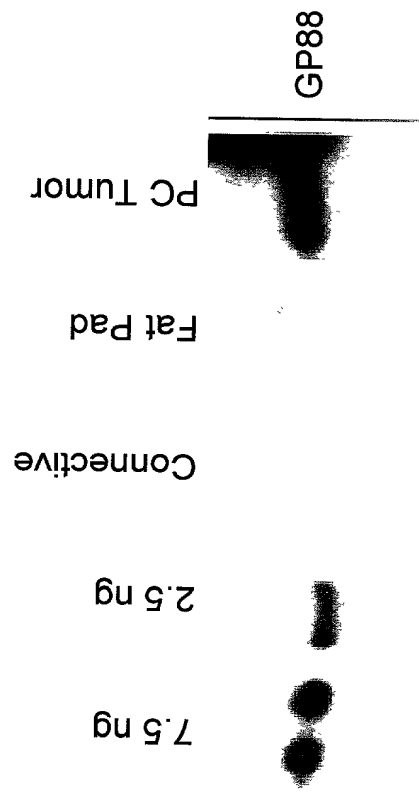


FIG.4

**GP88 mRNA EXPRESSION IN ESTROGEN-DEPENDENT AND
INDEPENDENT HUMAN MAMMARY CARCINOMA CELLS**



FIG. 5

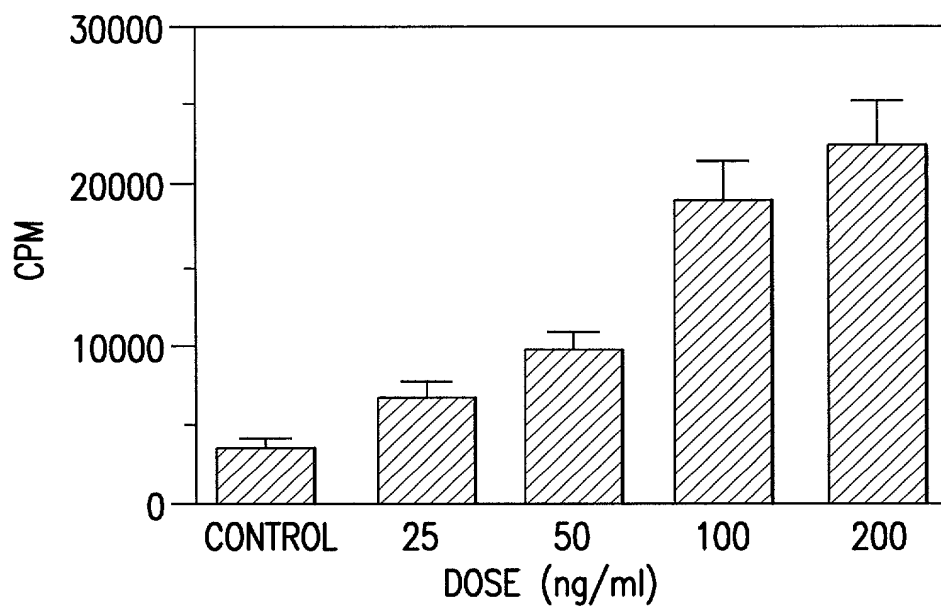


FIG. 6A

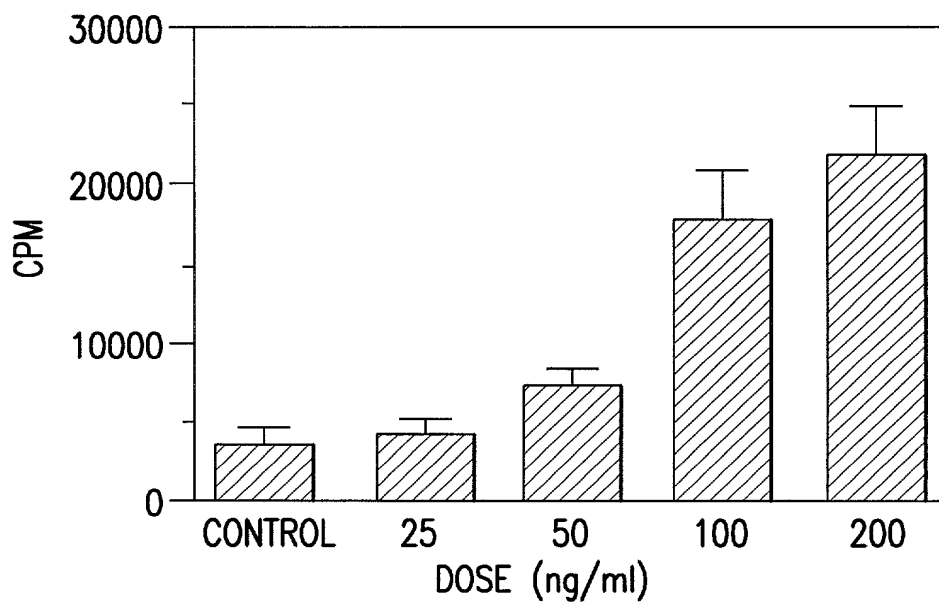


FIG. 6B

EXPRESSION OF GP88 IN ANTISENSE AND CONTROL TRANSFECTED PC CELLS



FIG.7

Mouse GP88 cDNA

CC	GGA	CCC	CGA	CGC	AGA	CAG	ACC	ATG	TGG	GTC	CTG	ATG	AGC	TGG	CTG	46
								M	W	V	L	M	S	W	L	8
GCC	TTC	GCG	GCA	GGG	CTG	GTA	GCC	GGA	ACA	CAG	TGT	CCA	GAT	GGG	CAG	94
A	F	A	A	G	L	V	A	G	T	Q	C	P	D	G	Q	24
TTC	TGC	CCT	GTT	GCC	TGC	TGC	CTT	GAC	CAG	GGA	GGA	GCC	AAC	TAC	AGC	142
F	C	P	V	A	C	C	L	D	Q	G	G	A	N	Y	S	40
TGC	TGT	AAC	CCT	CTT	CTG	GAC	ACA	TGG	CCT	AGA	ATA	ACG	AGC	CAT	CAT	190
C	C	N	P	L	L	D	T	W	P	R	I	T	S	H	H	56
CTA	GAT	GGC	TCC	TGC	CAG	ACC	CAT	GGC	CAC	TGT	CCT	GCT	GGC	TAT	TCT	238
L	D	G	S	C	Q	T	H	G	H	C	P	A	G	Y	S	72
TGT	CTT	CTC	ACT	GTG	TCT	GGG	ACT	TCC	AGC	TGC	TGC	CCG	TTC	TCT	AAG	286
C	L	L	T	V	S	G	T	S	S	C	C	P	F	S	K	88
GGT	GTG	TCT	TGT	GGT	GAT	GGC	TAC	CAC	TGC	TGC	CCC	CAG	GGC	TTC	CAC	334
G	V	S	C	G	D	G	Y	H	C	C	P	Q	G	F	H	104
TGT	AGT	GCA	GAT	GGG	AAA	TCC	TGC	TTC	CAG	ATG	TCA	GAT	AAC	CCC	TTG	382
C	S	A	D	G	K	S	C	F	Q	M	S	D	N	P	L	120
GGT	GCT	GTC	CAG	TGT	CCT	GGG	AGC	CAG	TTT	GAA	TGT	CCT	GAC	TCT	GCC	430
G	A	V	Q	C	P	G	S	Q	F	E	C	P	D	S	A	136
ACC	TGC	TGC	ATT	ATG	GTT	GAT	GGT	TCG	TGG	GGA	TGT	TGT	CCC	ATG	CCC	478
T	C	C	I	M	V	D	G	S	W	G	C	C	P	M	P	152
CAG	GCC	TCT	TGC	TGT	GAA	GAC	AGA	GTG	CAT	TGC	TGT	CCC	CAT	GGG	GCC	526
Q	A	S	C	C	E	D	R	V	H	C	C	P	H	G	A	168
TCC	TGT	GAC	CTG	GTT	CAC	ACA	CGA	TGC	GTT	TCA	CCC	ACG	GGC	ACC	CAC	574
S	C	D	L	V	H	T	R	C	V	S	P	T	G	T	H	184
ACC	CTA	CTA	AAG	AAG	TTC	CCT	GCA	CAA	AAG	ACC	AAC	AGG	GCA	GTG	TCT	622
T	L	L	K	K	F	P	A	Q	K	T	N	R	A	V	S	200
TTG	CCT	TTT	TCT	GTC	GTG	TGC	CCT	GAT	GCT	AAG	ACC	CAG	TGT	CCC	GAT	670
L	P	F	S	V	V	C	P	D	A	K	T	Q	C	P	D	216

FIG.8A

Mouse GP88 cDNA (continued)

GAT TCT ACC TGC TGT GAG CTA CCC ACT GGG AAG TAT GGC TGC TGT CCA	718
D S T C C E L P T G K Y G C C P	232
ATG CCC AAT GCC ATC TGC TGT TCC GAC CAC CTG CAC TGC TGC CCC CAG	766
M P N A I C C S D H L H C C P Q	248
GAC ACT GTA TGT GAC CTG ATC CAG AGT AAG TGC CTA TCC AAG AAC TAC	814
D T V C D L I Q S K C L S K N Y	264
ACC ACG GAT CTC CTG ACC AAG CTG CCT GGA TAC CCA GTG AAG GAG GTG	862
T T D L L T K L P G Y P V K E V	280
AAG TGC GAC ATG GAG GTG AGC TGC CCT GAA GGA TAT ACC TGC TGC CGC	910
K C D M E V S C P E G Y T C C R	296
CTC AAC ACT GGG GCC TGG GGC TGC TGT CCA TTT GCC AAG GCC GTG TGT	958
L N T G A W G C C P F A K A V C	312
TGT GAG GAT CAC ATT CAT TGC TGC CCG GCA GGG TTT CAG TGT CAC ACA	1006
C E D H I H C C P A G F Q C H T	328
GAG AAA GGA ACC TGC GAA ATG GGT ATC CTC CAA GTA CCC TGG ATG AAG	1054
E K G T C E X G I L Q V P W M <u>K</u>	344
AAG GTC ATA GCC CCC CTC CGC CTG CCA GAC CCA CAG ATC TTG AAG AGT	1102
<u>K V I A P L R L P D P Q I L K S</u>	360
GAT ACA CCT TGT GAT GAC TTC ACT AGG TGT CCT ACA AAC AAT ACC TGC	1150
<u>D T</u> P C D D F T R C P T N N T C	376
TGC AAA CTC AAT TCT GGG GAC TGG GGC TGC TGT CCC ATC CCA GAG GCT	1198
C K L N S G D W G C C P I P E A	392
GTC TGC TGC TCA GAC AAC CAG CAT TGC TGC CCT CAG GGC TTC ACA TGT	1246
V C C S D N Q H C C P Q G F T C	408
CTG GCT CAG GGG TAC TGT CAG AAG GGA GAC ACA ATG GTG GCT GGC CTG	1294
L A Q G Y C Q K G D T M V A G L	424
GAG AAG ATA CCT GCC CGC CAG ACA ACC CCG CTC CAA ATT GGA GAT ATC	1342
E K I P A R Q T T P L Q I G D I	440

FIG.8B

Mouse GP88 cDNA (continued)

GGT TGT GAC CAG CAT ACC AGC TGC CCA GTA GGG CAA ACC TGC TGC CCA	1390
G C D Q H T S C P V G Q T C C P	456
AGC CTC AAG GGA AGT TGG GCC TGC TGC CAG CTG CCC CAT GCT GTG TGC	1438
S L K G S W A C C Q L P H A V C	472
TGT GAG GAC CGG CAG CAC TGT TGC CCG GCC GGG TAC ACC TGC AAC GTG	1486
C E D R Q H C C P A G Y T C N V	488
AAG GCG AGG ACC TGT GAG AAG GAT GTC GAT TTT ATC CAG CCT CCC GTG	1534
K A R T C E K D V D F I Q P P V	504
CTC CTG ACC CTC GGC CCT AAG GTT GGG AAT GTG GAG TGT GGA GAA GGG	1582
L L T L G P K V G N V E C G E G	520
CAT TTC TGC CAT GAT AAC CAG ACC TGT TGT AAA GAC AGT GCA GGA GTC	1630
H F C H D N Q T C C K D S A G V	536
TGG GCC TGC TGT CCC TAC CTA AAG GGT GTC TGC TGT AGA GAT GGA CGT	1678
W A C C P Y L K G V C C R D G R	552
CAC TGT TGC CCC GGT GGC TTC CAC TGT TCA GCC AGG GGA ACC AAG TGT	1726
H C C P G G F H C <u>S A R G T K C</u>	568
TTG CGA AAG AAG ATT CCT CGC TGG GAC ATG TTT TTG AGG GAT CCG GTC	1774
<u>L R K K I P R</u> W D M F L R D P V	584
CCA ACA CCG CTA CTG TAA GGA AGG GCT ACA GAC TTA AGG AAC TCC ACA	1822
P R P L L *	589
GTC CTG GGA ACC CTG TTC CGA GGG TAC CCA CTA CTC AGG CCT CCC TAG	1870
CGC CTC CTC CCC TAA CGT CTC CCC GGC CTA CTC ATC CTG AGT CAC CCT	1918
ATC ACC ATG GGA GGT GGA GCC TCA AAC TAA AAC CTT CTT TTA TGG AAA	1966
GAA GGC TGT GGC CAA AAG CCC CGT ATC AAA CTG CCA TTT CTT CCG GTT	2014
TCT GTG GAC CTT GTG GCC AGG TGC TCT TCC CGA GCC ACA GGT GTT CTG	2062
<u>TGA GCT TGC TTG TGT GTG TGT GCG CGT GTG CGT GTG TTG CTC</u> <u>AAA TAA</u>	2110
<u>AGT</u> TTG TAC GCT TTC TGA AAA AAA AAA	2137

FIG.8C

Nucleotide sequence of human granulin/epithelin precursor (human GP88).
Human Granulin Genbank M75161\$

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[gcaggcaga ccatgtggac cttggtgagc tgggtggcct taacagcagg gctggtggct
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gggaggtggg gcctcaatct aaggcccttc cctgtcagaa ggggggttgg gcaaaagccc
attacaagct gccatcccct cccggtttca gtggaccctg tggccagggt cttttcccta
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FIG.9A

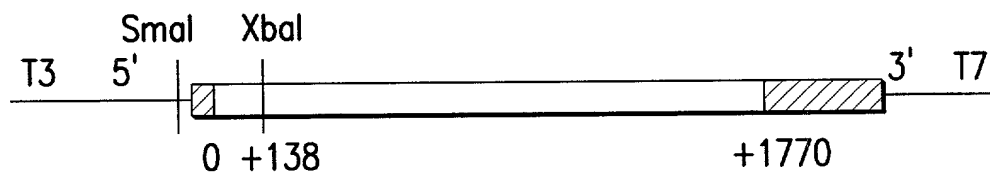
Amino-acid sequence of human granulin/epithelin precursor (human GP88).

MWTLVSWVALTAGLVAGTRCPDGQFCPVACCLDPGGASYSCRP
LLDKWPTTLSRHLGGPCQVDAHCSAGHSCIFTVSGTSSCCPFPEAVACGDGHHCCPRG
FHCSADGRSCFQRSGNNSVGAIQCPDSQFECPDFSTCCVMVDGSWGCCPMPQASCCED
RVHCCPHGAFCDLVHTRCITPTGTHPLAKKLPARTNRAVALSSSVMCPDARSRCPDG
STCCELPSGKYGCCPMPNATCCSDHLHCCPQDTVCDLIQSKCLSKENATTDLLTYLPA
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CEQGPHQVPWMEKAPAHLSLPDPQALKRDVPCDNVSSCPSSDTCCQLTSGEWGCCPIP
EAVCCSDHQHCCPQRYTCVAEGQCQRGSEIVAGLEKMPARRGSLSHPRDIGCDQHTSC
PVGGTCCPSQGGSWACCQLPHAVCCEDRQHCCPAGYTCNVKARSCEKEVVSAQPATFL
ARSPHVGKDVCEGEGHFCHDNQTCCRDNRQGWACCPYAQGVCCADRRHCCPAGFRCA
RRGTKCLRREAPRWDAPLRDPALRQLL*

FIG.9B

FIG. 10

GP88 cDNA CLONE in SK



STRUCTURE OF pCMV₄ EXPRESSION VECTOR

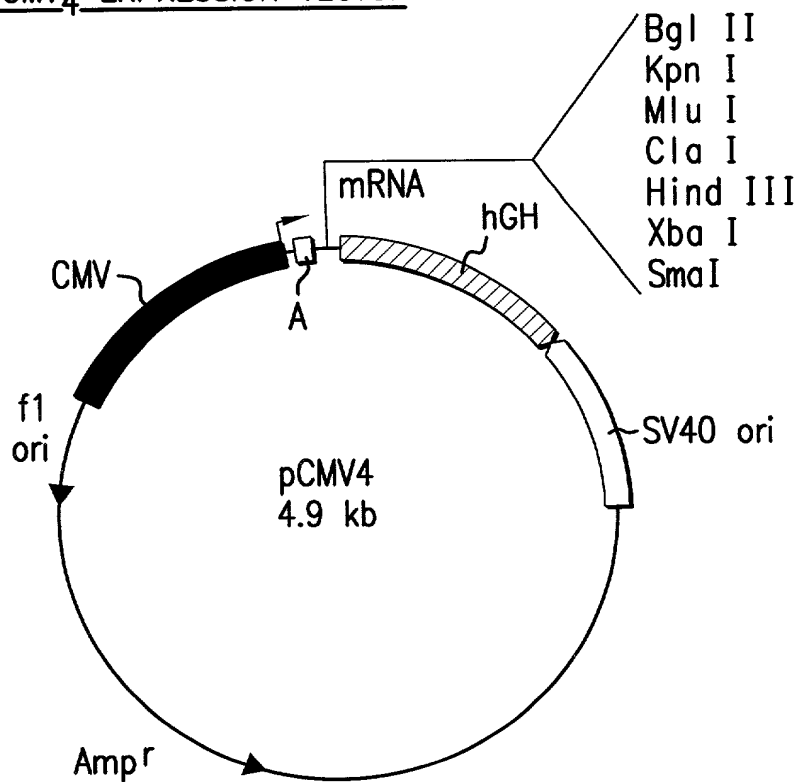


FIG.11

CROSS-LINKING OF ^{125}I -rGP88 TO CCL64 CELLS

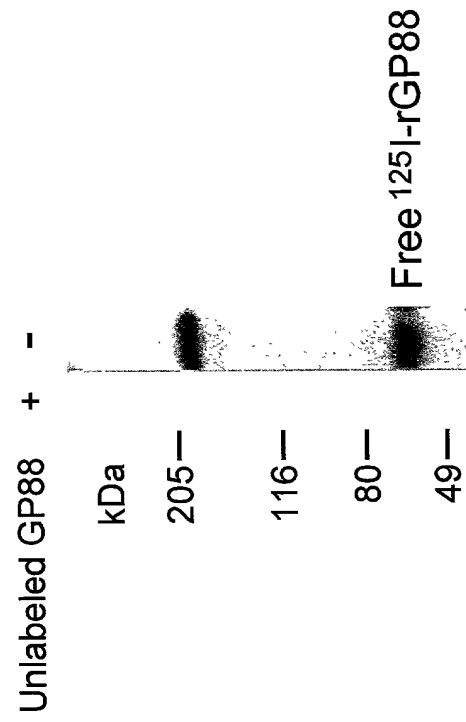


FIG.12

CROSS-LINKING OF ¹²⁵I-rGP88

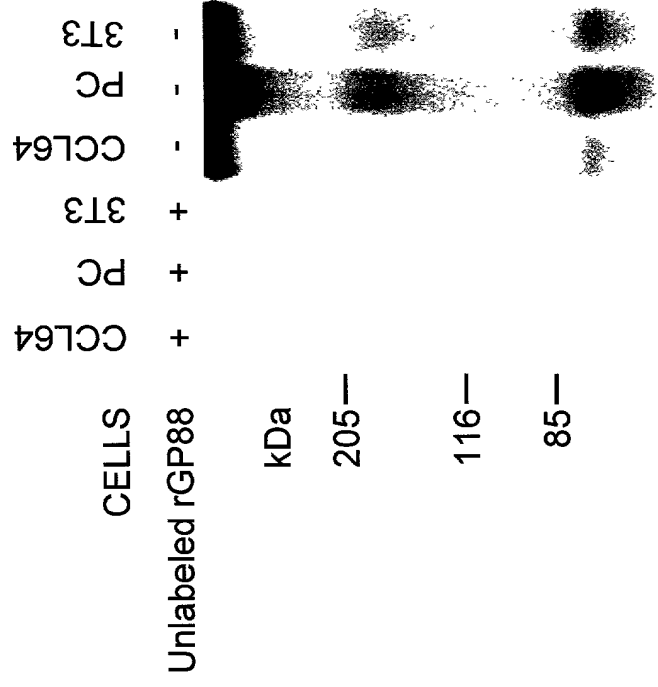


FIG.13

**GP88 EXPRESSION IN NON TUMORIGENIC (MCF 10A)
AND MALIGNANT (MCF 7, MDA-468) HUMAN
MAMMARY EPITHELIAL CELLS**

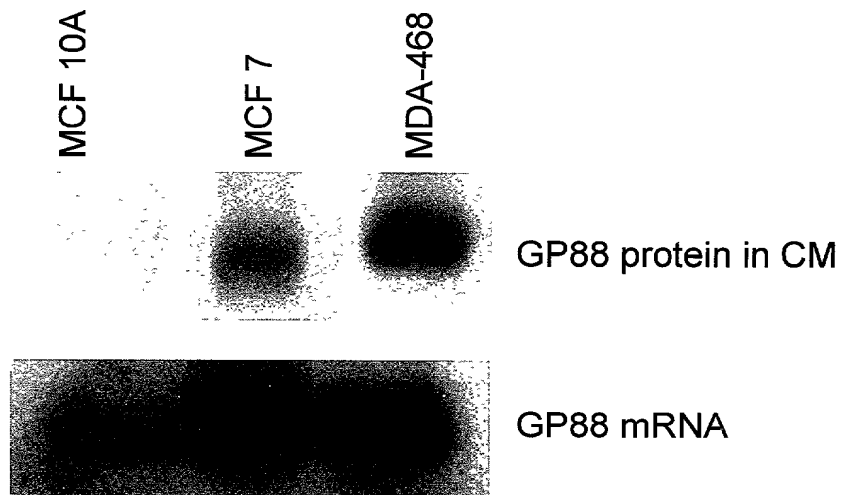


FIG.14

**GP88 EXPRESSION IS INHIBITED BY ANTISENSE GP88
cDNA TRANSFECTION IN HUMAN BREAST
CARCINOMA MDA-468**

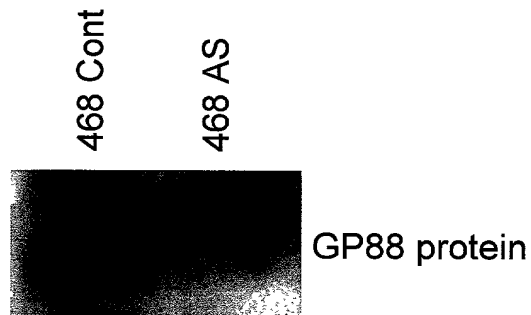
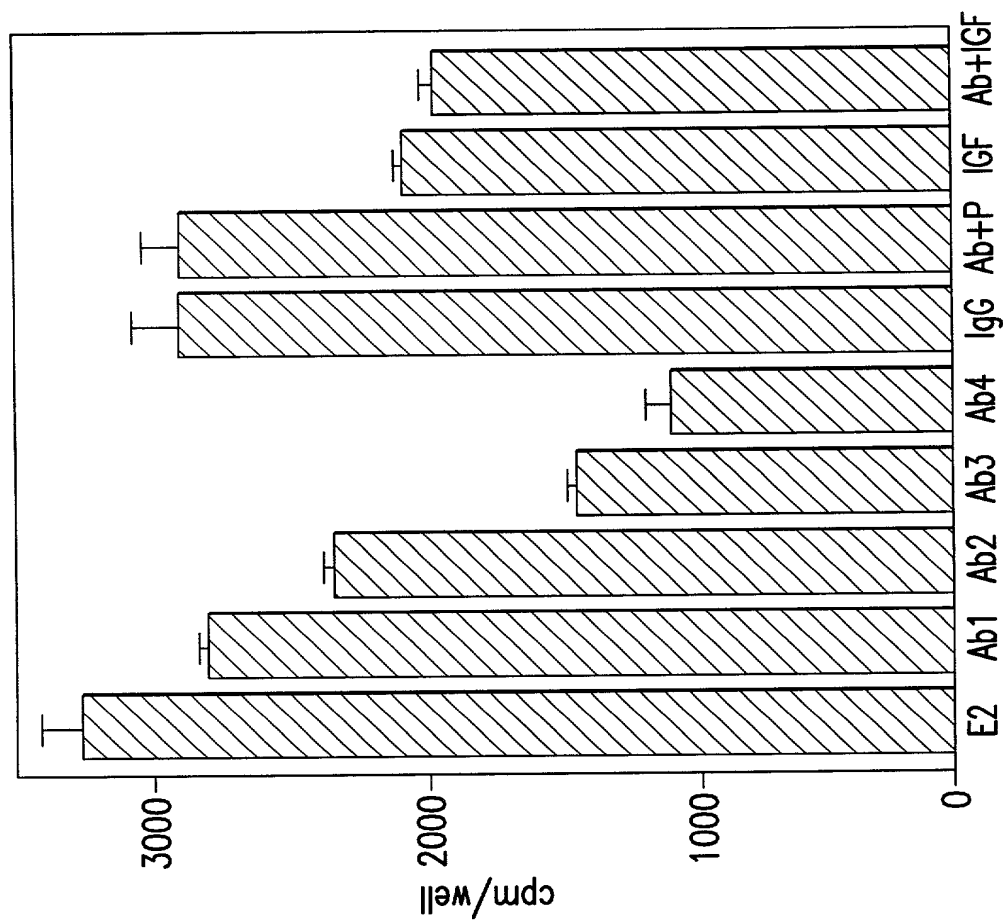
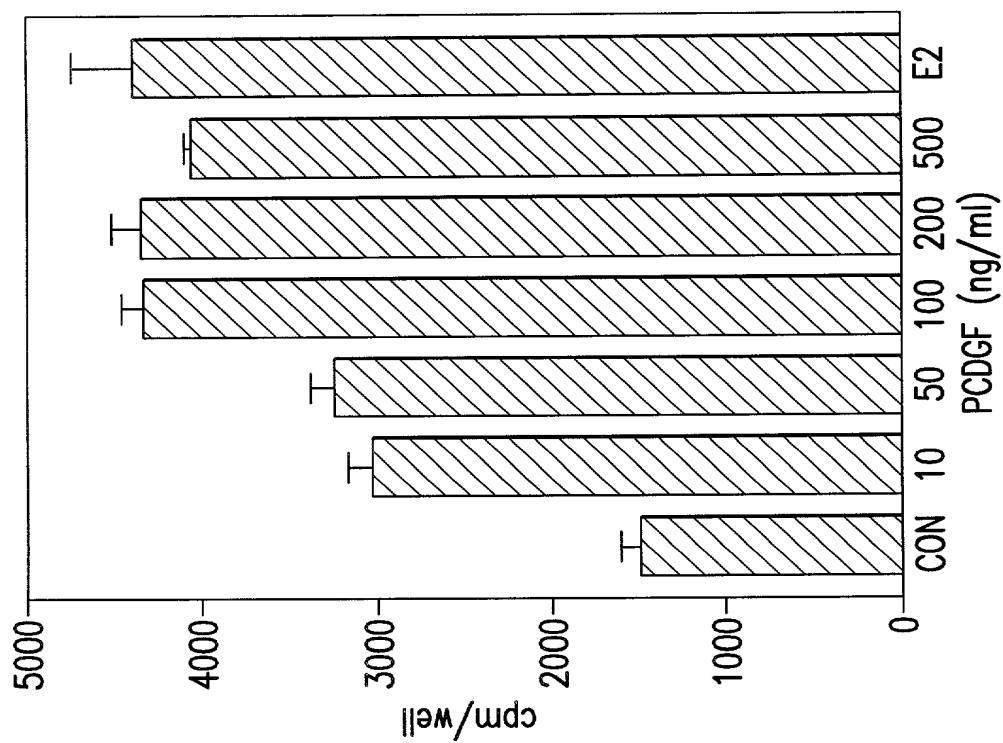


FIG.15



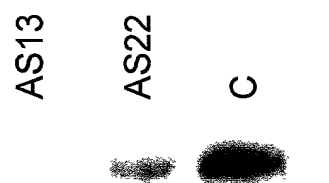


FIG.17A

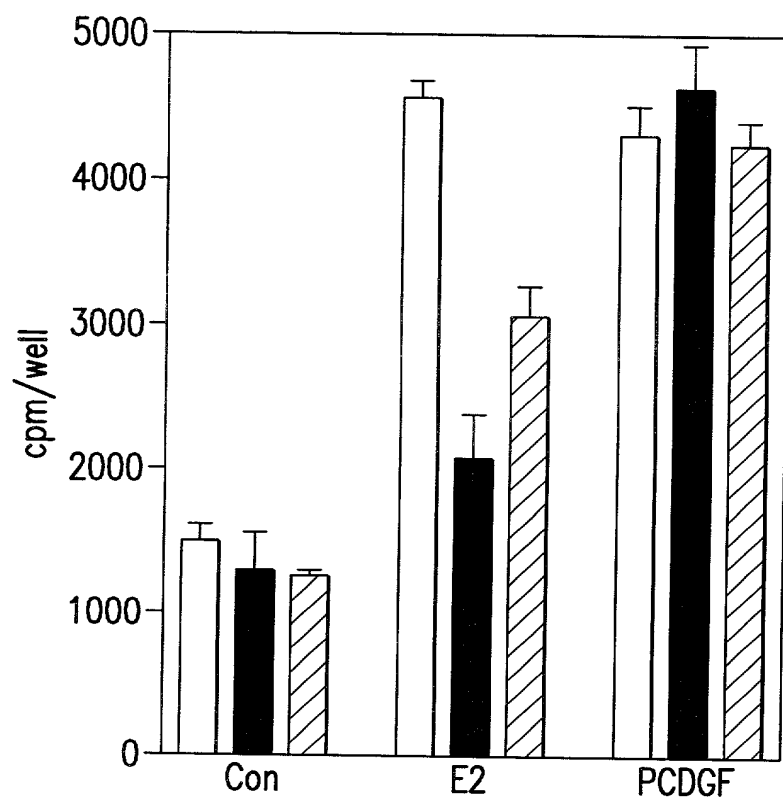


FIG.17B

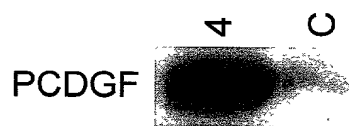


FIG.18A

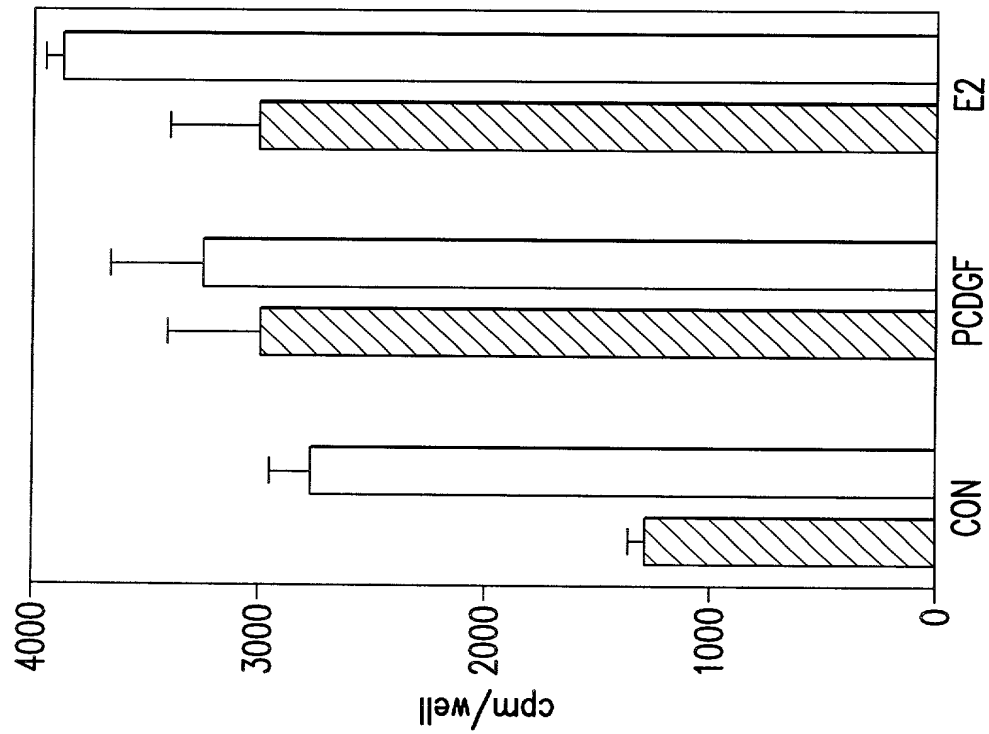


FIG.18B

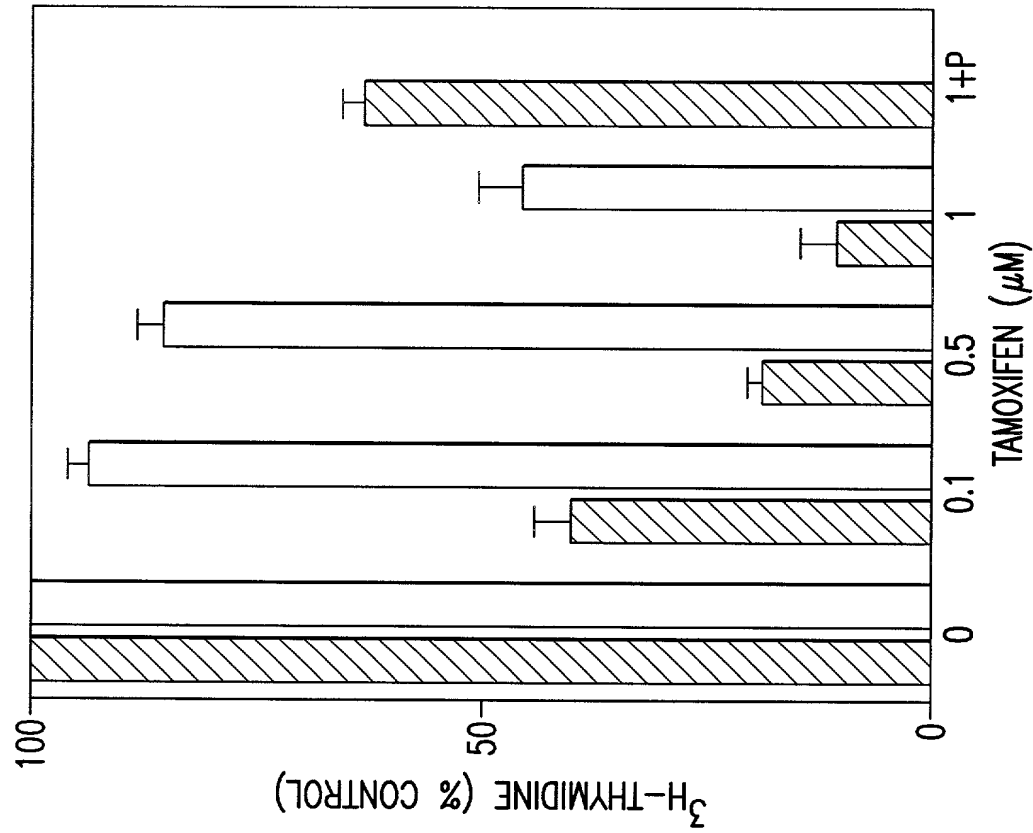


FIG.18C

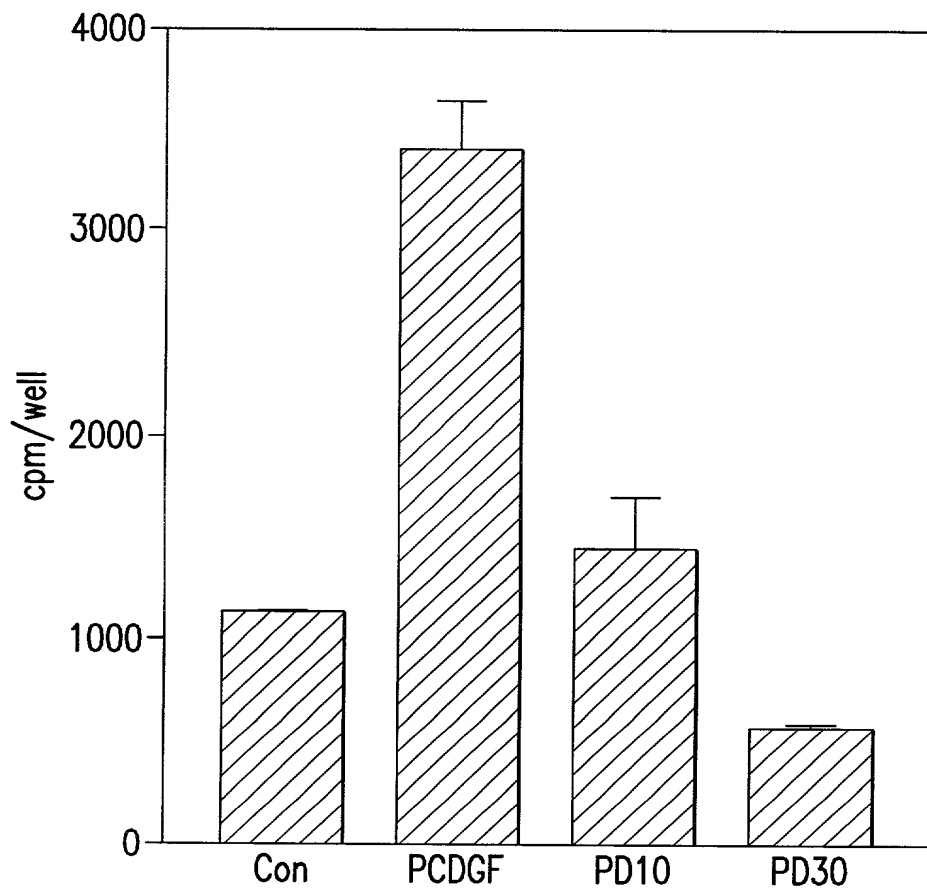


FIG.19A

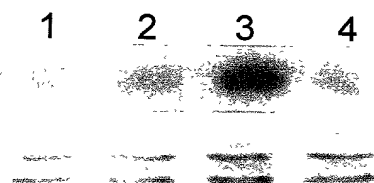


FIG.19B

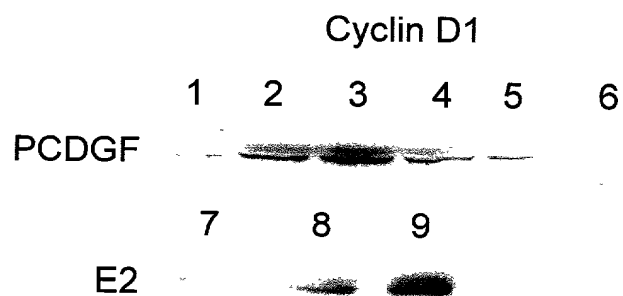


FIG.20A

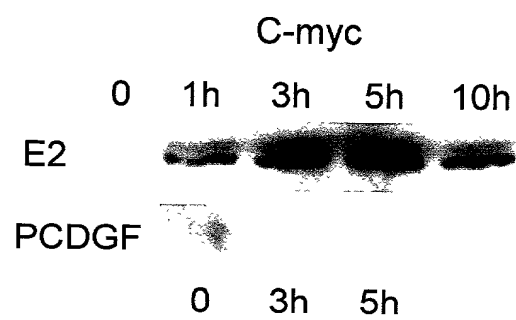
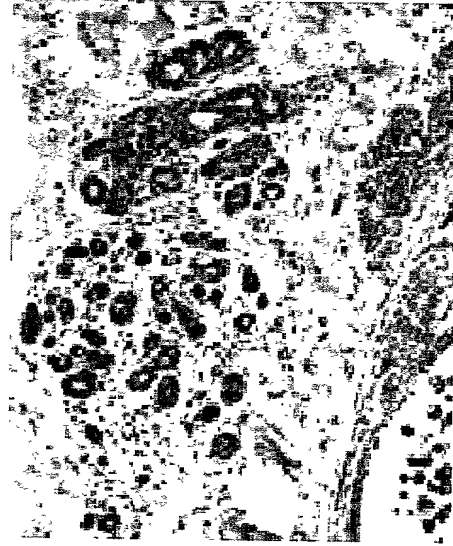


FIG.20B



BENIGN LESION



DUCTAL INVASIVE CARCINOMA

GP88 STAINING WITH ANTI-GP88 ANTIBODY
ON PARAFFIN EMBEDDED BREAST CANCER BIOPSIES
BY IMMUNOHISTOCHEMISTRY (IHC)

FIG.21

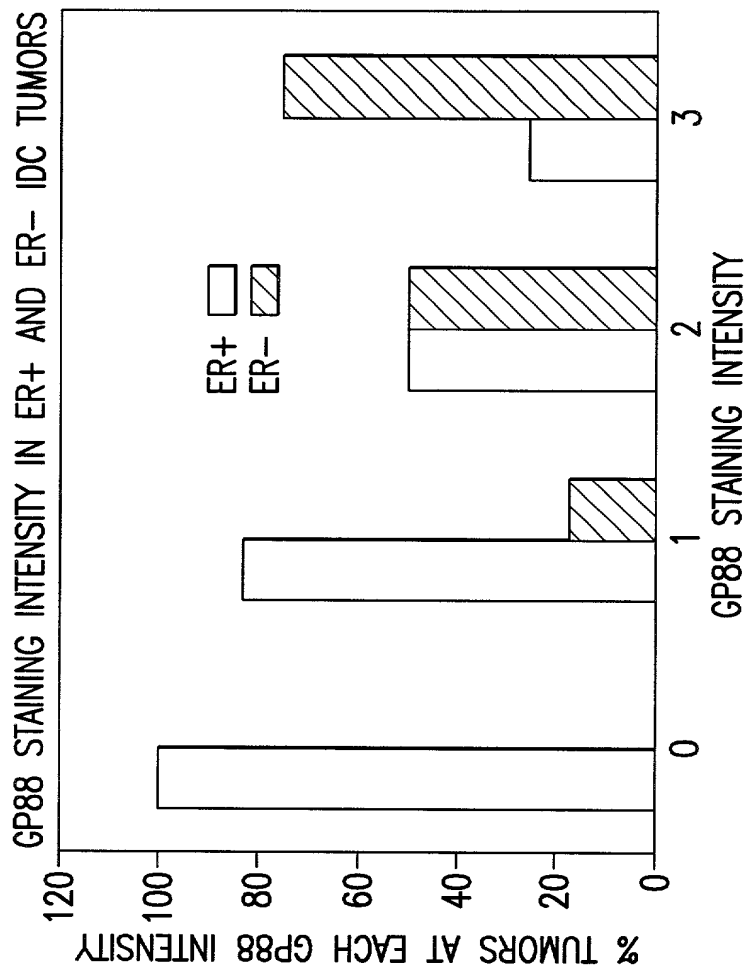


FIG.22

TAMOXIFEN RESPONSIVENESS OF ER+ IDC TUMORS CLASSIFIED BY GP88 STAINING INTENSITY

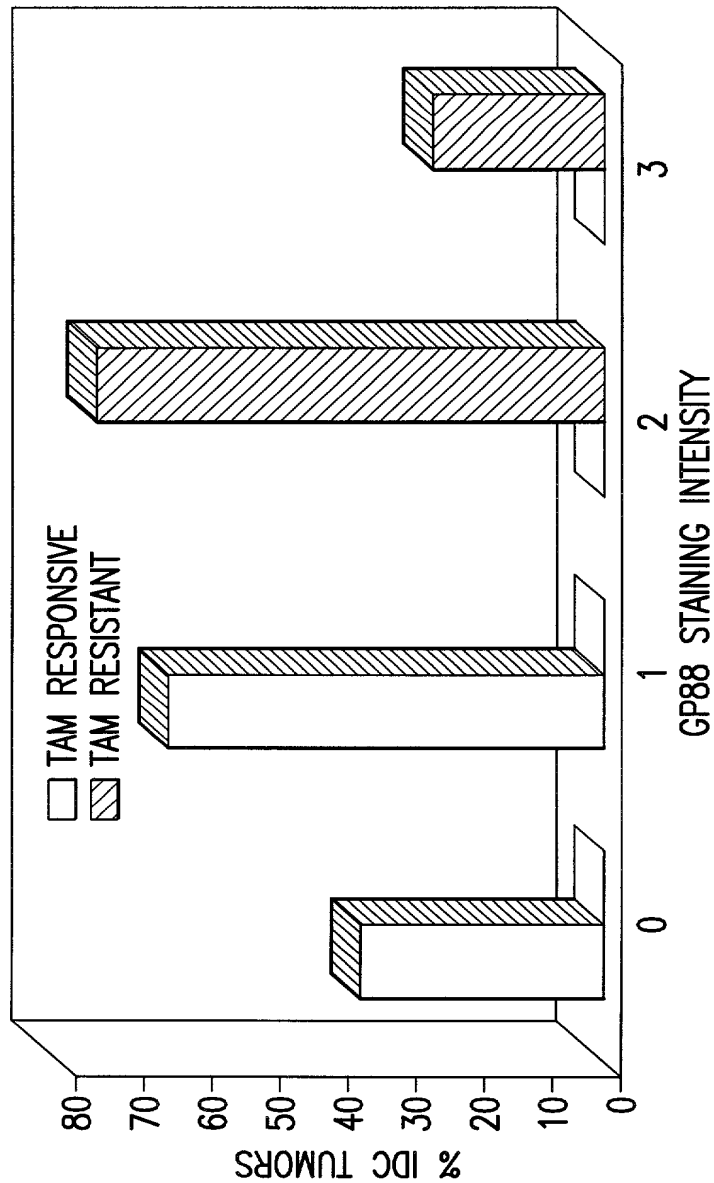


FIG.23